

# POLLINATION ECOLOGY OF *CASSIA ALATA* L. (CAESALPINIACEAE)<sup>1</sup>

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(With one text-figure)

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*Cassia alata* blooms annually during October-February. The flowers are borne on typical racemes, and are yellow and devoid of nectar. They exhibit enantiostyly and heteranthery with feeding as well as pollinating anthers. In the biotope of the study area, the carpenter bees consisting of *Xylocopa latipes* and *X. pubescens* are the exclusive visitors to *C. alata*. They collect pollen by buzzing. During the vibratile movement of the visitor's body, the pollen grains are discharged from pollinating anthers on to the sides of the insect visitor's thorax and abdomen. Simultaneously, the pollen grains are also transferred to the stigmas oriented to the right or to the left. Heteranthery and enantiostyly, complemented by the buzzing behaviour of the pollinating carpenter bees, promote geitonogamy as well as xenogamy in *C. alata*.

## INTRODUCTION

Buchmann (1983) gave a complete list of anthecological work on *Cassia* species which include *C. fasciculata*, *C. fistula*, *C. glauca*, *C. bacillaris*, *C. multijuga*, *C. didymobotrya*, *C. auriculata*, and *C. alata*. Those works not mentioned by Buchmann (1983) have been touched upon by Gottsberger and Gottsberger (1988). These studies show that pollen discharge by release of a small cloud of pollen occurs from apical pores of the anthers by vibratile behaviour of the bees collecting pollen. The genus *Cassia* is known to exhibit heteranthery and enantiostyly. Heteranthery means that the androecium of the same flower is functionally differentiated into short stamens with feeding anthers, which provide food for the pollinator, and longer stamens with pollinating anthers which provide pollen for pollinating the stigma. Enantiostyly refers to the occurrence of right-

styled and left-styled flowers on the same inflorescence. This floral dimorphism has been treated as a device for enhancing outbreeding (Bahadur *et al.* 1990). However, Dulberger (1981) cautions that it need not be connected with outcrossing.

Carpenter bees are the only insects pollinating *Cassia* sp. so far studied. The bees collect pollen from feeding anthers by rapidly contracting the indirect flight muscles, thus producing strong vibrations that are transmitted directly to the anthers and indicated by audible buzzing of the bees. This vibration rapidly produces a directed stream or pollen cloud from the anther pores that primarily strikes the venter of the bee, and sometimes the pleural and dorsal areas also. During this act, the pollen from pollinating anthers situated over the dorsal side or lateral sides of the bee is discharged and deposited on the head-thorax region of the foraging bee, resulting in nototriby or pleurotriby. The stigma of the flower simultaneously strikes the pollen-laden head-thorax region of the bee.

Pijl (1954) has provided fragmentary information on the floral biology of *C. alata* in Java. He suggests that *C. alata* has a similar

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floral mechanism and is pollinated by carpenter bees. The available information on this medicinally valuable plant is highly inadequate to understand its unique form of pollination, and the information is from Java, where meteorological conditions may differ from those of India. Complete details of the floral ecology of *C. alata* are necessary for its commercial cultivation.

#### MATERIAL AND METHODS

*Cassia alata* L. is abundant in Visakhapatnam (17° 42' N and 82° 18' E). The studies were carried out in 1989 and 1990. To determine flowering phenology, ten inflorescences selected at random were tagged before the initiation of blooming and observed for the daily opening of flowers. The inflorescences were then removed to avoid recounting the next day. Tagged inflorescences were observed until they ceased flowering to obtain data on the duration of flower production and also the daily rate of flowering. Flower morphometrics were observed in detail. Daily anthesis rate was noted from ten randomly selected inflorescences marked before anthesis. Time of anther dehiscence was recorded by observing the anthers with a 10x hand lens before and after the flowers opened. Undehisced mature anthers, immersed separately in a drop of lactophenol aniline-blue were observed under the microscope, and the number of pollen grains per anther was counted to determine pollen output. Pollen production per anther was then multiplied by the number of anthers to estimate total pollen grains per flower. Structural characters of pollen grains were observed and their size was measured with a calibrated ocular micrometer. Pollen-ovule ratio was determined by dividing the number of pollen grains per flower by the number of ovules per flower. Pollen viability was tested by hand-pollination experiments using relatively fresh stigmas, and stigma receptivity by hand-pollination using relatively fresh pollen.

Breeding behaviour was studied through controlled pollination. Fruit set, seed set and fecundity rates in controlled pollination were measured following the procedure described by Aluri and Subba Reddi (1994). The flower and fruit abortions were expressed in percentages.

Flower visitors were carpenter bees of two species of *Xylocopa* only. Foraging activity of these bees, their foraging behaviour, forage resource sought, pollination potential, etc. was investigated in detail. The pollen carrying capacity of the bees was also determined by counting the pollen grains obtained from body washings in aniline-blue. The duration of flower visit and flowers foraged in a unit time by the flower-visitors were noted to assess the foraging speed.

#### RESULTS

**Flowering phenology:** The plants of *C. alata* grow from new seeds every year following the first rains of the monsoon and continue to grow until late September. Then they start producing inflorescence stalks and flower buds. The mature flower buds begin to open from October through February. A plant flowers for an average period of 88 days (range 61-106).

**Inflorescence phenology:** The inflorescence is a typical terminal raceme. An inflorescence produces a mean number of 88 flowers (range 40-117) anthesing over an average period of 24 days (range 16-29). Each day the number of newly opened flowers of an inflorescence varies from 2-5. Flower production ceases in February, then the plants start to dry up and wither.

**Flower morphology:** The flowers are bisexual, showy but lack fragrance. The calyx consists of 5 sepals, each 1.5 cm long. The yellow corolla is ovate and divided into 5 petals. The clawed and imbricated petals are first involute and later become spread. Stamens are 10. The uppermost (adaxial) three stamens have sterile anthers. The remainder possess fertile anthers.

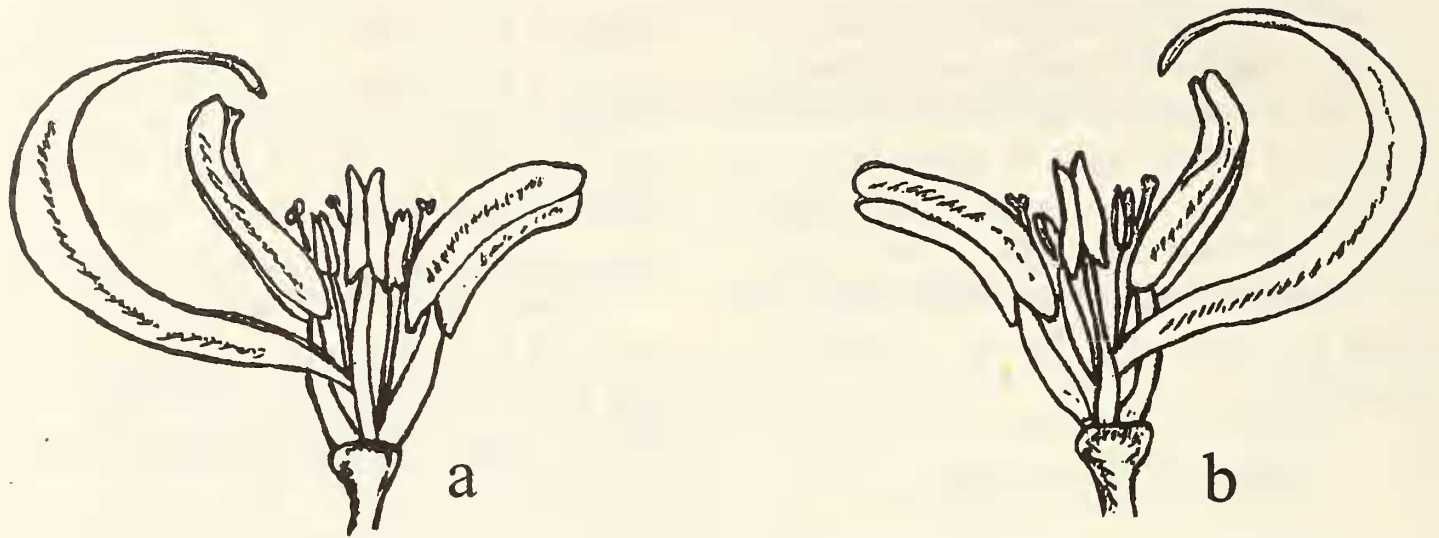


Fig. 1: Enantiostyly in *Cassia alata* L. a. Left-styled flower; b. Right-styled flower

which can be grouped into three categories. The top central group of 4 stamens have filaments 3 mm long and anthers 4 mm long. The second group composed of 2 large lateral stamens having filaments 4 mm long and anthers 10 mm long, are situated below the top central group. The anthers of this group are thick, hard, curved, tapering and slightly twisted. The third group represents one lower-most (abaxial) median stamen with a 7 mm long filament and a 5 mm long anther. The ovary has 60 ovules (range 50-63) arranged in a linear fashion. The upwardly bending sickle-shaped pistil emerges between the bases of the second group of stamens and above the median stamen of the third group. The pistil projects to the right or to the left in the flowers of the same inflorescence (Fig. 1), but without any regular pattern. The ratio of right-styled and left-styled flowers in an inflorescence is nearly 1:1. The style is terminated with a simple stigma having a cavity-like opening.

**Anthesis and anther dehiscence:** The flowers open daily during 0300-0400 h. Anthers dehisce by pores; a pore is formed at the apical appendage of each anther lobe. Dehiscence takes place about 3 h after anthesis. The corolla, together with stamens, falls off in 36-48 h after anthesis.

**Pollen characters:** Pollen grains are triangular, tricolporate, with a smooth exine. The grains are 43-51  $\mu\text{m}$  in diameter. Pollen

production per anther in the three categories of fertile stamens is different. It averages 32,600 (range 29,000-37,000) in the top central group of stamens. The corresponding figures for the second and the third group of stamens is 2,47,000 (range 2,10,640-2,86,000) and 22,410 (range 18,550-25,460) respectively. A few grains are sterile in all the three groups of stamens (3-5%). The pollen-ovule ratio is 9,880:1.

**Pollen viability and stigma receptivity:** *C. alata* pollen germinated well in 50% sugar concentration. The germination percentage obtained for fresh pollen is 90%, 24 h old pollen 68% and for 100 h old pollen 6%. The pollen viability tested through fruiting ability showed that the stigma pollinated with fresh pollen produced 84% and those with 96 h old pollen gave 20% fruiting.

The tests conducted for fruiting ability of the stigmas of different maturing periods from anthesis to the flower life up to 36 h showed that the stigma favours pollen germination and later gives fruit for 30 h. This condition of the stigma is taken as stigma receptivity period. The fruiting ability of the stigma, however, was not the same throughout the receptivity period. The percentage of fruiting obtained with fresh stigmas was 84%. With advancement in the age of the stigma, there was reduction in fruiting, it being 12% with 30 h old stigmas.

**Breeding behaviour:** The results of breeding tests show that the plant is not apomictic, but reproduces through xenogamy and geitonogamy. The xenogamic fruiting, seeding and fecundity values obtained were 84,100 and 84%, respectively. The agreeing values for geitonogamy were 68,100 and 68%.

**Natural flower and fruit abortions:** The recorded natural flower and fruiting pattern show that about half of the flowers fall off without any signs of fruit development. In the remainder, about 25% initiate fruit development and then abort. The remaining flowers ranging from 19-25% develop mature fruit. The mature fruits are largely produced at the basal part of the inflorescence.

**Flower visitors:** The flowers of *C. alata* are foraged exclusively by *Xylocopa pubescens* and *X. latipes*. The foraging visits of these bees are, however, significantly disproportionate. *X. latipes* made 64-70% of foraging visits while *X. pubescens* only 30-36%. But the two species were equally mobile. Both foraged 13 flowers (range 7-20) in one minute and spent 2-4 seconds at a flower. The bees usually foraged on fresh flowers and occasionally on day-old flowers. The foraging speed of the two species was greater on sunny days compared to cloudy days. On sunny days, the bees started foraging at 0800 h and stopped at 1600 h. The activity was more brisk around noon. The foraging schedule of the bees was delayed by two hours and stopped early by an hour on cloudy days.

**Foraging behaviour:** The yellow corolla appears to attract and direct carpenter bees to the pollen source in the flower. On approaching the flower, the bee lands on the top central and the third group of stamens situated in the middle of the flower. While landing, it curls its body over the anthers and produces a high-pitched buzzing sound which is quite different from the flight sound. The head and thorax of the buzzing bee are now covered over by the upper involute petals, while the lower petals are pressed downward by the weight of the bee. The bee

vibrates its body and collects pollen from the top central and third group of anthers. A buzzing sound is heard and the pollen is deposited all over the ventral side of the bee, where it is greatly accessible for grooming and for subsequent ingestion. Added to this, the stigma does not make contact with the ventral side of the bee. The four stamens of the top central group and the single stamen of the third group constitute the feeding anthers. The second group, with larger stamens oriented laterally, discharge pollen onto the sides of the thorax and abdomen of the bee. Washings of the thorax revealed 180 to 325 pollen grains, while those of the abdomen area showed 180-260 grains. The pollen deposited in this area may be totally inaccessible for grooming or for ingestion. The stigma of the right- and left-styled flowers touches the dorsal side of the thorax and the abdomen, where pollen from lateral anthers is deposited. The second category of stamens, the lateral ones; thus constitute the pollinating anthers.

#### DISCUSSION

As early as 1909, Burkill considered *Xylocopa* sp. to be the most important flower-visiting insects in the plains of tropical India, and largely responsible for the pollination of sunhemp, Indian pulses and Cassias. From other tropical regions *Xylocopa* sp. have been reported as pollinators of several *Cassia* species. Knuth (1906) reported that in South Asia, the main pollinators of *Cassia* spp. with large and medium sized flowers are *X. latipes* and *X. aestuans*. Pijl (1954) describes three large-flowered tropical species of *Cassia* with typical *Xylocopa* flowers; one of these species is *Cassia alata*. We observed that the flowers of *C. alata* in Visakhapatnam are visited and pollinated exclusively by *X. latipes* and *X. pubescens*.

*C. alata* flowers are nectarless, and the floral reward is only pollen. The male carpenter bees do not forage for pollen (Buchmann, 1983). Then successful pollination in *C. alata* should

result from the activity of female carpenter bees. The captured pollinators in the present study were female.

On the basis of P/O ratio (9880:1), *C. alata* may be treated as highly xenogamous (Cruden, 1977). However, breeding experiments revealed that both xenogamy and geitonogamy operate with equal success in seed formation. Cruden and Jensen (1979) suggest that P/O ratios may reflect pollination efficiency, and the lower P/O ratio makes more efficient delivery of pollen. Thus, in *C. alata* the high P/O ratio may indicate inefficient delivery of pollen.

However, the 100% seed set and the nototribic and/or pleurotribic transfer of pollen suggest that the pollination mechanism in *C. alata* is efficient. The high pollen production which resulted in the high P/O ratio compensates the pollen wastage/loss from pollinator feeding. Further, the small size of the stigma, compared to the area of pollen spread on the body of the bee, may also necessitate the production of large amounts of pollen. These interpretations agree with those of Dulberger (1981) who also found unusually high P/O in *C. didymobotrya* and *C. auriculata*.

Flower function and bee behaviour suggest that geitonogamy cannot be eliminated. When the bee lands on an inflorescence, it visits all the fresh flowers that range from 2-5. In every visit of the bee, its dorsal thorax or abdomen, powdered with pollen grains makes contact with the stigma. Then, the pollen grains are transferred to the stigma. The pollen thus transferred may be geitonogamous or xenogamous.

Contrary to the present observations, Pijl (1954) observed in Java that isolated plants of *C. alata* are sterile. He writes that the flowers of an old specimen of *C. alata* have been setting fruit to almost 100%. After clearing the surrounding vegetation in the whole field no fruit was formed. As soon as the neighbouring young plant, which had also been spared, started flowering, the inflorescence of the first one produced fruits again. Perhaps, *C. alata* may

be having different races in different geographical regions, with differing breeding systems.

Heteranthery and enantiostyly in *Cassia* species might be taken as a device for promoting outcrossing (Bahadur *et al.* 1990). Efficacy of this strategy in promoting outcrossing in *C. alata* is poor. The flowers have their pollinating anthers oriented on both sides of the flower and the stigma, whether in R and L flowers, can invariably receive pollen of the two pollinating anthers. Since the flowers are compatible to both geitonogamous and xenogamous pollen, and the pollinator visits all the 2-5 flowers that open in a day, the situation may enhance geitonogamous pollination, irrespective of the presence of enantiostyly. However, considering the flight pattern of carpenter bees which involve interplant and interpopulation flights, it is likely that xenogamous pollination is also promoted, though the incidence of such pollination relative to geitonogamy can only be assessed through pollen tagging tests. Dulberger (1981), while interpreting the functions of various morphological characters of *C. auriculata* and *C. didymobotrya*, concluded that the main function of enantiostyly need not be connected with outcrossing. He says that its primary role may be that of clearing access of the insect to the feeding anthers, at the same time protecting the female parts from injury by insect vibrations. Possible injury may be prevented by the presentation of stigma and style from the median plane of symmetry, so that it comes into contact with the back or side of the insect rather than its ventral part. As revealed by the extensive studies of vibratile pollination (Buchmann, 1983), the intensity of buzzing increases with the size of the insect. Carpenter bees are the giants of the bee world (Meeuse 1961) and are to be treated as powerful buzzers (Roubik, 1989). Therefore, it is reasonable to assume that in flowers exhibiting the buzz-pollination syndrome discussed at length by Buchmann (1983), the style deflections may have evolved together with

changes in stamen orientation and a division of androecium into upper and lower anthers. It may be noted that the female part deflected away from the flower centre is the whole carpel. Enantiostyly might be a safeguarding device from the damage that may result from the weight of the pollinator and intensity of its vibrations.

Gottsberger and Gottsberger (1988) state that in their study on the flower adaptation and evolution of the Cassinae in relation to pollination events, there was often no real contact of the bee body with the stigma and/or anther openings, but that there occurred close approximation. Then, to explain the deposition of pollen on the stigma, they relied on the concept of Corbet *et al.* (1982) that electrostatic forces play an important role in pollen transfer via insects. Electrostatic potentials enable pollen to jump from anther to bee and from bee to stigma.

Even if there is direct contact, as seen in the present study of *C. alata*, electrostatic forces are necessary to push or pull pollen into the hollow style tip wherein the stigmatic surface is located. As stated by Buchmann (1983) and Gottsberger and Gottsberger (1988), there would be a lot of pollen dispersal and loss into the air but for the electrostatic field around a visiting bee. In the absence of electrostatic field around the pollination vector, the cloud of pollen released during vibration should become airborne and pollen trapping in the atmosphere should reveal high concentrations. However, pollen trapping in the ambient air has revealed low concentrations of *Cassia* pollen. The probable importance of electrostatic forces in buzz-pollination has also been recognised by Buchmann (1978) and Buchmann and Hurley (1978).

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